## THE ABSORPTION OF RADIOISOTOPES BY CERTAIN MICROORGANISMS 1, 2

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The safe disposal of radioactive wastes is one of the major problems associated with the use of atomic energy. High level wastes are generally produced in small volumes making it possible to keep this material segregated from the environment. Before high level wastes are buried or stored they are concentrated and encased in some nonpermeable material. Although the disposal of these high level radioactive wastes is expensive they offer no threat to the general population.

Low level radioactive wastes, in contrast, are now being produced in large volumes and in the near future will amount to many billions of gallons yearly. Because it is economically and physically impossible to concentrate these wastes so that they can be disposed of in a manner similar to that used for high level wastes, the only alternative is to release them into the environment. When radioactive elements are permitted to enter the air, soil and water there is a tendency for plants and animals to utilize these chemicals in their own metabolic processes. In many instances radioactive isotopes are accumulated by microorganisms and enter into the food chain. If a particular species of radioactive element is involved in the metabolism of plants or animals, it may be concentrated many thousands of times over the concentration present in the ambient media.

Measuring and predicting the uptake and accumulation of radionuclides by microorganisms presents many problems. Results obtained in the laboratory are not translatable to natural conditions but can be used as a comparison among organisms. Unfortunately laboratory methods involve the use of specific media for specific organisms which is contradictory to conditions prevailing in nature. This laboratory has found that a number of other conditions affect the uptake of radionuclides and must remain constant to obtain usable data. The conditions investigated are as follows:

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- 1. Temperature was found to greatly influence the rate of radionuclide uptake by microorganisms. A temperature of  $75 \pm 1^{\circ}$ F was selected for all data present.
- 2. The size and shape of the vessel containing the culture also affected the uptake. The ratio of glass surface to the volume of medium as well as the type of glass influence reactions involved. Wide mouth Pyrex flasks, 500 ml., were employed for all cultures.
- 3. Even culture conditions at different laboratories vary. For example, a formula for a particular media may call for spring water or sea water. Within the State of Florida the total solids content of spring water varies from less than 100 mg/1 to greater than 25,000 mg/1 and so-called sea water from the West coast to the East coast may vary almost as much. Sea water from two sources and spring water from one source were used in my experiments but, the comparison of data from different laboratories would be greatly facilitated if the basic solvent of all media was distilled water. Three media were adapted for this investigation. Chlamydomonas, Platymonas and Nitzschia were grown in Rice's medium with sea water. Bacteria were cultured in Zobell's medium. Guillard's medium prepared with sea water was used for Rhodomonas and Guillard's medium prepared with spring water supported the growth of Navicula. Ochromonas were grown in wheat medium.
- 4. The control of pH was found to be very difficult. It was observed that at the end of 24 hours twenty replicate cultures exhibited twenty different pH values with a spread of 2.5 pH units. This shows the need for a strong buffer system which is lacking in many culture media for protozoa and algae.
- 5. Forty foot-candles of light was provided by Sylvania 48T12 cool white fluorescent lamps. It was found by experimentation that light from 20 foot-candles to 80 foot-candles did not appreciably affect the uptake of radioisotopes.
- 6. The age and concentration of cultures also strongly affected the uptake rate of radioisotopes. The cultures employed in this investigation were about 1 month old and were all still in the active growth cycle.

The accumulation of radioisotopes was determined by first adding a measured amount of a specific radionuclide to the culture. After a given incubation time, usually 48 hours, the organisms were filtered from the medium, dried at  $103^{\circ}$ F and weighed. The concentration of radioactivity in the microorganisms expressed in micromicrocuries per milligram,  $\mu\mu c/mg$ , of dried organisms divided by the original concentration of radioactivity in  $\mu\mu c/mg$  of medium was taken as the concentration factor. Results are presented in Tables 1-7.

TABLE 1 THE UPTAKE OF RADIOISOTOPES BY OCHROMONAS SP.

Radioisotope	Initial Conc. in Medium μμc/mg	Incubation time hours	Concentration Factor	Deviation
Cerium-141	0.37	48	15,170	± 7.0%
Cesium-137	0.21	48	960	$\pm$ 9.3%
	0.52	48	1,120	$\pm$ 8.4%
	242	48	1,130	$\pm$ 17.4%
Cobalt-60	0.50	48	1,070	$\pm 74.9\%$
	2.20	48	1,500	$\pm 24.3\%$
	4.15	48	1,160	$\pm 19.3\%$
Copper-64	1.22	48	3,040	$\pm 11.8\%$
	6.16	48	1,960	$\pm 26.7\%$
	26.6	48	1,840	$\pm$ 6.9%
Iron-59	0.04	48	1,550	± 86 %
	0.25	48	2,700	$\pm 126$ %
	0.55	48	4,480	$\pm 43.7\%$
Mixed Fission	0.3	48	16,800	$\pm 47.3\%$
Products	0.6	48	15,500	$\pm 40.2\%$
Niobium-95	0.04	48	25,500	$\pm 11.6\%$
	0.5	48	34,900	$\pm 19.7\%$
Phosphorus-32	1.3	48	10,500	$\pm 16.8\%$
(PO <sub>4</sub> )	4.0	48	4,900	$\pm 29.1\%$
Ruthenium-103	0.25	48	4,000	$\pm 10.2\%$
	0.57	48	6,500	$\pm 11.7\%$
Strontium-89	5.5	48	3,060	$\pm$ 9.2%
Uranium-238* (UO <sub>2</sub> )	6.1	48	330	$\pm$ 4.2%
Tungsten-185	23	48	20	± 4.2%
$(WO_i)$	102	48	29	± 5.3%
,	182	48	20.6	± 2.2%
	383	48	9.1	$\pm 19.3\%$
Yttrium-91	2.9	48	46,600	± 29.2%
Zinc-65	1.0	48	7,600	± 11.2%
	2.4	48	6,900	$\pm$ 5.1%

<sup>\*</sup>Natural uranium

TABLE 2 THE UPTAKE OF RADIOISOTOPES BY PLATYMONAS SP.

Radioisotope	Initial Conc. in Medium μμc/mg	Incubation time hours	Concentration Factor	Deviation
Cerium-141	4.8	48	5,100	± 17.5%
Cesium-137	0.20	48	150	$\pm 19.1\%$
	0.40	48	121	$\pm 11.2\%$
	1.0	48	50	$\pm 21.1\%$
	5.0	48	36	$\pm 58.3\%$
Iron-59	0.05	48	720	$\pm 14.9\%$
	0.20	48	1,030	$\pm 21.6\%$
	0.60	48	950	$\pm 17.7\%$
Mixed Fission	0.20	48	11,000	$\pm 43.5\%$
Products	0.45	48	13,300	$\pm 38.4\%$
	0.80	48	16,000	$\pm 37.5\%$
Phosphorus-32	2.0	48	6,300	$\pm 14.3\%$
$(PO_4)$	3.0	48	7,500	$\pm 16.1\%$
	4.0	48	13,600	$\pm 49.2\%$
Promethium-147	6.0	48	4,500	± 6.8%
	1.0	48	5,200	$\pm 11.2\%$
Praseodymium-142	1.8	48	3,800	$\pm 16.8\%$
Strontium-89	0.40	48	457	$\pm 21.6\%$
	0.79	48	652	$\pm 26.9\%$
Yttrium-91	1.3	48	13,500	$\pm 43.5\%$
Zinc-65	0.40	48	53,800	$\pm 12.5\%$
	0.62	48	67,800	$\pm 17.3\%$

TABLE 3 THE UPTAKE OF RADIOISOTOPES BY NAVICULA CONFERVACEA

Radioisotopes	Initial Conc. in Medium	Incubation time hours	Concentration Factor	Deviation
Cesium-137	3.5	48	2,180	± 33.9%
Cobalt-60	1.0	48	. 271	$\pm 16.3\%$
Iron-59	1.5	48	4,020	$\pm 12.7\%$
Mixed Fission				
Products	0.75	48	12,500	$\pm 27.6\%$
Niobium-95	0.4	48	83,700	$\pm 31.1\%$
Promethium-147	3.2	48	2,160	$\pm$ 7.5%
Ruthenium-103	1.1	48	7,900	$\pm 27.4\%$
Strontium-89	0.20	48	1,380	$\pm$ 7.2%
Yttrium-91	0.71	48	2,230	$\pm$ 64.4%
Zinc-65	0.50	48	23,600	$\pm 15.3\%$

TABLE 4 THE UPTAKE OF RADIOISOTOPES BY CHLAMYDOMONAS SP.

Radioisotopes	Initial Conc. in Medium	Deviation		
Cerium-141	0.50	48	7,400	± 24.4%
Cesium-137	0.89	48	28.7	$\pm 11.3\%$
Iron-59	1.25	48	6,000	$\pm 16.8\%$
Mixed Fission				
Products	0.75	48	16,800	$\pm 27.5\%$
Promethium-147	0.60	48	12,600	$\pm 47.8\%$
Strontium-89	0.85	48	1,000	$\pm 15.2\%$
Zinc-65	2.0	48	20,000	$\pm$ 26.7%

TABLE 5 THE UPTAKE OF RADIOISOTOPES BY NITZSCHIA SP.

Radioisotope	Initial Conc.	Incubation time hours	Concentration Factor	Deviation
Cerium-141	1.0	48	28,500	± 19.3%
Cesium-137	0.65	48	97	$\pm$ 2.1%
Iron-59	0.30	48	5,533	$\pm 26.9\%$
Mixed Fission				
Products	0.55	48	11,500	$\pm 47.3\%$
Promethium-147	0.6	48	2,420	$\pm 11.3\%$
Strontium-89	0.85	48	650	$\pm 31.8\%$
Zinc-65	0.52	48	42,000	$\pm$ 17.3%

TABLE 6 THE UPTAKE OF RADIOISOTOPES BY RHODOMONAS SP.

Radioisotope	Initial Conc. in Medium	Incubation time hours	Concentration Factor	Deviation
Cesium-137	4.2	48	36	± 14.5%
Iron-59	0.30	48	7,500	$\pm 36.7\%$
Mixed Fission				
Products	0.85	48	1,700	$\pm 23.3\%$
Promethium-147	0.70	48	10,000	$\pm$ 63.2%
Strontium-89	2.0	48	100	$\pm 14.6\%$
Zinc-65	1.7	48	312	$\pm 35.5\%$

TABLE 7

THE UPTAKE OF RADIOISOTOPES BY VARIOUS BACTERIA

Organism	Radioisotope	Initial Conc. in Medium	Incubation time hours	Concentration Factor	Deviation
Flavobacterium					
Aquatile	Cesium-137	0.23	24	26	$\pm 17.4\%$
Sphaerotilus sp.	Cesium-137	0.20	24	116	$\pm 11.3\%$
Sphaerotilus sp.	Copper-64	0.93	24	3,890	$\pm 46.5\%$
Zooglea ramigera	Cesium-137	0.24	24	558	$\pm 21.7\%$
Marine Bacteria'					
Z-5	Yttrium-91	0.15	24	886	$\pm 33.9\%$
Z-7	Promethium-147	0.55	24	310	$\pm 25.9\%$
Z-8	Strontium-89	1.40	24	104	$\pm 8.9\%$
Z-8	Cesium-137	0.73	24	15	$\pm 49.1\%$
Z-9	Strontium-89	1.40	24	100	$\pm 11.3\%$
Z-19	Copper-64	1.37	24	990	$\pm 32.6\%$
Z-20	Zinc-65	0.94	24	290	$\pm 15.6\%$
Z-21	Promethium-147	0.60	24	147	$\pm 8.2\%$
Z-21	Cerium-141	0.64	24	280	$\pm 11.3\%$
Z-22	Cerium-141	0.94	24	1,740	$\pm 61.2\%$

## RESULTS AND CONCLUSIONS

Most of the trivalent rare earth elements are markedly concentrated by microorganisms. This group of elements is important because of its relative abundance in fission products. It is extremely doubtful whether or not these elements are required for metabolic processes. It is more likely that these radioisotopes are absorbed and/or ingested after which the colloidal state is formed. This would render the elimination of the element more difficult.

The uptake of strontium-89 is relatively low when compared to the trivalent rare earth elements which indicates that the microorganisms tested have a low calcium requirement. A concentration factor of about 20,000 for strontium was observed in several diatoms that use calcium.

Zinc-75 was accumulated extensively by all of the organisms except *Rhodomonas* and bacteria. Yttrium and iron isotopes were markedly concentrated. Cesium, like strontium, was not concentrated to any great extent by the microorganisms tested.

The concentration factors given in Tables 1-7 are of value primarily as a comparison between the microorganisms tested. These factors are calculated on a dry weight basis for the organisms and not on a wet weight basis because it was thought that these conditions were more reproducible. The low accumulation of strontium and cesium does not in any way lessen the hazardous nature of these elements. Animals of the second trophic level not only feed on phytoplankton but are also capable of concentrating these elements directly. This then does not eliminate the biologically hazardous radioisotopes from the food chain. The marked uptake of the rare earth elements indicates that following a nuclear detonation or reactor accident a high degree of radioactive pollution of fish and shellfish would be expected. This group of radioisotopes decays at a fairly rapid rate which, along with the loss of radioactivity by cell division, would decrease the critical concentration of the rare earth elements in second and third trophic levels. Additional research and data are needed before parameters can be established for the levels of radioactivity in phytoplankton which will indicate critical contamination to man's food.